IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant: Cyrus Rustam Kumana and Yok-Lam Kwong

Serial No.: 10/669,869 Art Unit: 1616

Filed: September 23, 2003 Examiner: Frank I. Choi

For: *FORMULATION OF ORAL COMPOSITIONS COMRISING ARSENIC TRIOXIDE AND METHODS OF USE THEREOF*

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

DECLARATION UNDER 37 C.F.R. § 1.132

Sir:

We the undersigned, Cyrus Rustam Kumana, B.Sc., M.B.B.S., F.R.C.P. [Canada, London, Edinburgh, Glasgow] and Yok-Lam Kwong, M.D., F.R.C.P. (Edin), F.R.C.Path (U.K.) do hereby declare and state that:

1. We are co-inventors of the above-identified application.

2. I, Cyrus Rustam Kumana, am currently Professor Emeritus in the Department of Medicine, at The University of Hong Kong, and an Executive Editor of the Hong Kong Medical Journal. Till July 2004, I was the Founding Chair Professor of Clinical Pharmacology & Therapeutics, in the Department of Medicine, at The University of Hong Kong. I am a medical doctor, specialized in the field of medical therapeutics, which includes the interaction between drugs and the human body in health and disease.

3. I, Yok-Lam Kwong, am currently a Chair Professor in the Department of Medicine, at The University of Hong Kong, working in the team of haematology, haematological oncology and bone marrow transplantation. I am a medical doctor specializing in the field of haematology, haematological oncology and bone marrow transplantation.

4. We have read the Office action mailed on February 7, 2007 in connection with the above-identified application and the references cited therein.

5. The claims of the above-identified application refer to a dosage formulation for oral administration containing an effective amount to treat a person in need thereof of arsenic trioxide when administered orally. The claims further specify that the amount of arsenic trioxide in the oral dosage formulation is less than the effective amount for intravenous administration. Dependent claims further specify that the dose is 5-10 mg/day, or in the form of tablets or other dosage forms which are clearly not administerable intravenously. The claims also refer to a method of treating hematological malignancies in a subject in need thereof. The method includes orally administering to the subject a therapeutically effective amount of arsenic trioxide with fewer side effects than the same amount of arsenic trioxide administered intravenously.

6. We understand that the Examiner cited WO 99/24029 and CN1370540 in his novelty and obviousness rejections of the claims.

7. Aresenic trioxide blocks potassium currents I_{Kr} and I_{Ks}, causing QT prolongation. The QT interval represents the time for both ventricular depolarization and repolarization to occur, and therefore estimates the duration of an average ventricular action potential. Prolongation of QT or corrected QT (QTc) increases the risks of ventricular

tachyarrhythmias. QT-interval dispersion measures regional nonhomogeneities of ventricular repolarization. Greater dispersions increase ventricular arrhythmias. Ventricular tachyarrhythmias are reported in about 30% of patients treated with intravenous As_2O_3. The following experiments demonstrate the surprisingly fewer dangerous cardiac side effects associated with oral administration of arsenic trioxide when compared with intravenous administration.

8. **Materials and Methods**

Patients

Seventeen consecutive patients with relapsed acute promyelocytic leukemia APL (Table 1) were studied. All had normal left ventricular ejection fraction and blood biochemistry.

Table 1. Characteristics and baseline laboratory data of 17 patients with relapsed APL treated with As_2O_3

Sex/Age	LVEF	Previous Treatment	Creatinine	Na^+	K^+	Ca^{2+}	Status*	Outcome+
F/40	55%	ATRA, Dauno	81	141	3.7	2.41	CR	CR, 22 m+
F/49	78%	ATRA, Dauno, VP-16, Ara-C, IDA	85	142	3.5	2.47	CR	CR, 85 m+
F/35	75%	ATRA, Dauno, VP-16, Ara-C, IDA	59	139	4.2	2.39	CR	CR, 49 m+
F/32	65%	ATRA, Dauno, VP-16	82	141	3.8	2.46	CR	CR, 26 m+
M/41	55%	ATRA, Dauno, Ara-C	89	139	2.8	2.39	CR	CR, 55 m+
M/76	50%	ATRA, Dauno	146	135	4.1	2.48	CR	Sepsis, 15 days
M/46	65%	Duano, VP-16	87	144	3.5	2.25	CR	CR, 26 m+
F/55	50%	IDA	87	143	3.4	2.43	CR	CR, 21 m+
M/48	50%	ATRA, Dauno, VP-16, Ara-C	100	140	3.6	2.38	CR	CR, 40 m+
M/64	50%	ATRA, 5:2, Ara-C, IDA	143	141	4.3	2.27	CR	CR, 49 m+
F/50	50%	Dauno, VP-16, Ara-C	100	137	3.9	2.30	CR	CR, 65 m+
M/37	50%	ATRA, Dauno, VP-16, Ara-C	78	140	3.8	2.40	CR	Dead, R3, 16 m
M/40	65%	ATRA, Dauno, VP-16, Ara-C, IDA	126	135	3.7	2.33	CR	CR, 15 m+
M/44	55%	ATRA, Dauno, VP-16, Ara-C	73	141	3.6	2.36	CR	Dead, TB, 12 m
M/44	50%	ATRA, Dauno, VP-16	68	142	3.6	2.38	CR	CR, 31 m+
M/28	75%	ATRA, Dauno, VP-16	73	141	3.8	2.38	CR	CR, 11 m+
M/31	65%	ATRA, Dauno, VP-16, Ara-C	88	146	4.1	2.43	CR	CR, 22 m+

LVEF indicates left ventricular ejection fraction; F, female; ATRA, all-*trans* retinoic acid; Dauno, daunorubicin; CR, complete remission; Ara-C, cytosine arabinoside; VP-16, etoposide; IDA, idarubicin; M, male; R3, third relapse; TB, tuberculosis.

*Status after completion of the first course of oral As_2O_3.

[+]Status at latest follow-up, with maintenance therapy comprising oral As_2O_3 (10 mg/d) and ATRA (45 mg/m2/d), given for 2 weeks every 2 months for a planned 2 years.

Study protocol

Patients received oral arsenic trioxide (As_2O_3) (10 mg/d) for 2 weeks, followed by a drug-free period of 6 to 8 weeks before the next course. Patients took oral As_2O_3 at 14:00 every day to match the timing for electrocardiography (ECG) and Holter recording, and blood arsenic assay. The treatment protocol was approved by the institutional review board of Queen Mary Hospital, and all patients gave their informed consent in accordance with the Declaration of Helsinki.

ECG and Holter measurement

Data were collected at day 10 of oral As_2O_3 (As_2O_3-ON) and 4 weeks after stopping oral As_2O_3 (As_2O_3-OFF). Resting 12-lead surface ECGs were recorded (paper-speed: 50 mm/second) for measuring QT intervals. QT intervals at each lead and the corresponding RR-interval at lead II were measured to calculate the corrected QT (QTc) (Bazett formula: QTc = QT/square-root of RR interval). QT dispersion was determined as the difference between the maximum and minimal QT measured from each of 12 leads. During As_2O_3-ON and As_2O_3-OFF periods, 24-hour Holter monitoring (Zymed DigiTrak Plus; Zymed 1810, Philips, The Netherlands) was performed to assess circadian QT variations.

Data interpretation

Holter recordings were reviewed and edited manually. Recordings had to exceed 20 hours and be of good quality to be analyzed. The time domain (standard deviation of normal RR interval [SDNN]) and frequency domain (ratio of low-frequency to high-

frequency power [LF/HF]) of HVR was measured to assess parasympathetic and sympathetic activities, respectively. Sixteen ECG and Holter recordings were analyzed by a cardiologist blinded to patient treatment.

Elemental arsenic levels

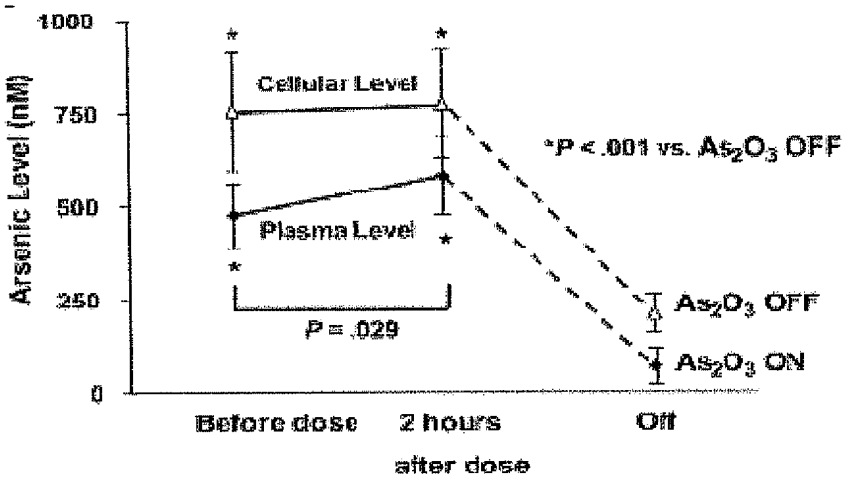
After venepuncture, EDTA-anticoagulated blood was immediately separated into plasma and cell fractions. Arsenic levels were measured by inductively coupled plasma mass spectrometry.

Statistical analysis

Continuous variables were expressed as mean plus or minus standard error of the mean. Comparisons were performed with the Student *t* test or Fisher exact test (SPSS software, version 10.0). *P* values less than .05 were considered significant.

9. The following results were obtained using the methodology described above.

10. Consistently, cellular arsenic levels were significantly higher than plasma arsenic levels as demonstrated in the graph below. At steady state, during As_2O_3-ON, an oral dose of As_2O_3 (10 mg) did not cause a change in cellular arsenic level. Four weeks after cessation of treatment (As_2O_3-OFF), there was a near complete clearance of arsenic.



11. Both QT and QTc were significantly longer during As_2O_3-ON than in As_2O_3-OFF (Table 2, $P < .01$). However, the QT and QTc dispersion were comparable during As_2O_3-ON and As_2O_3-OFF (Table 2, $P > .05$).

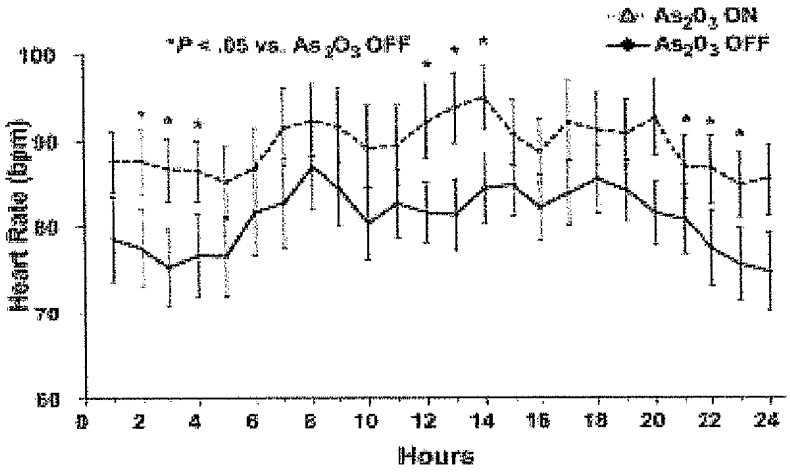
Table 2. Changes in QT intervals and heart rate variability during oral As_2O_3 therapy

12-lead ECG	As_2O_3-ON	As_2O_3-OFF	p values
QT (milliseconds)	383 ± 8	359 ± 8	0.08
QTc (milliseconds)	455 ± 6	423 ± 7	< 0.01
QT dispersion	54 ± 4	54 ± 4	0.82
QTc dispersion	65 ± 5	62 ± 6	0.78
Holter recording			
SDNN (milliseconds)	90 ± 11	118 ± 12	0.02
LF (normalized unit)	26 ± 2	26 ± 2	0.97
HF (normalized unit)	22 ± 3	24 ± 3	0.21
LF/HF ratio	1.36 ± 80.16	1.45 ± 0.16	0.30

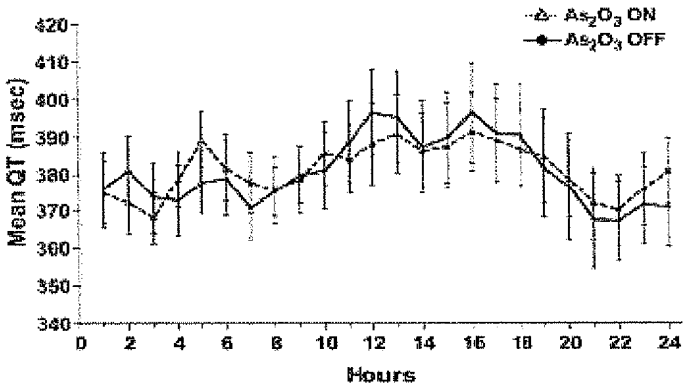
For the purposes of this table, P values greater than .05 are not significant.

LF, low frequency; HF indicates high frequency; SDNN, standard deviation of normal to normal RR interval.

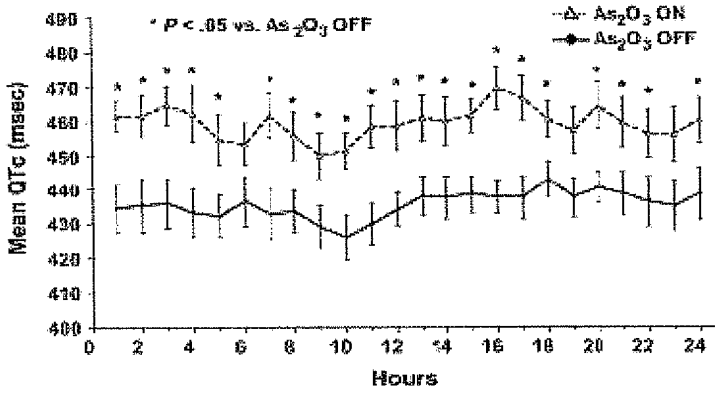
12. Holter monitoring showed similar circadian heart rate variations during As_2O_3-ON and As_2O_3-OFF, but 24-hour heart rates were consistently higher during As_2O_3-ON than in As_2O_3-OFF, being significantly different during late evening (21:00, 22:00, 23:00), early morning (02:00, 03:00, 04;00), and afternoon (12:00, 13:00, 14:00; $P < .05$) as shown in the graph below.



13. As shown in the graph below, QT intervals over 24 hours showed marked variations, although the pattern and measurement at each hour were comparable between As_2O_3-ON and As_2O_3-OFF.



14. Circadian variations of mean QTc intervals were comparable between As_2O_3-ON and As_2O_3-OFF, but measurements at each hour were significantly longer during As_2O_3-ON ($P < .05$, except at 06:00, 19:00, 23:00) as shown in the graph below.



15. The mean differences of QTc between As_2O_3-ON and As_2O_3-OFF are shown in the graph below. QTc prolongation of more than 30 milliseconds was observed at only one time point (16:00, 2 hours after oral As2O3). No case had QTc prolongation of more than 50 milliseconds. This resulted in a QTc of more than 500 milliseconds in only 3 of 16 patients, all within 4 hours of oral As2O3.

16. Table 2 shows that although the SDNN was significantly lower during As_2O_3-ON than in As_2O_3-OFF, there was no significant difference in LF/HF between As_2O_3-ON and As_2O_3-OFF.

17. Ventricular premature beats were comparably frequent between As_2O_3-ON and As_2O_3-OFF (0.03% ± 0.02% versus 0.03% ± 0.03%; $P = 0.8$). No ventricular tachyarrhythmia was observed.

18. These results demonstrate that, when measuring for indicators of proarrhythmic risks associated with oral arsenic trioxide: significant QTc prolongation greater than 30

milliseconds was only observed at a single-time-point (2 hours post-oral arsenic trioxide composition), QTc prolongation never exceeded 50 milliseconds, and QTc intervals greater than 500 milliseconds were only observed in three patients within four hours of oral arsenic trioxide composition. Importantly, these observations showed virtually no ventricular proarrythmia in all patients studied. More importantly, none of the patients experienced ventricular tachyarrhythmias. These unexpected results are vastly superior to patients who receive intravenous arsenic trioxide compositions, where 26% of patients have QT intervals greater than or equal to 500 milliseconds, with QTc intervals prolonged by 30-60 milliseconds in 36.6% of treatment courses, and by QTc intervals prolonged by more than 60 milliseconds in 35.4% of patients, resulting in ventricular tachyarrhythmias (more specifically, torsades de pointes) in 1% of cases.

19. One of ordinary skill in the art would not have been able to predict in view of the references cited by the Examiner that the claimed arsenic trioxide formulation is orally bioavailable at substantially the same amount as the intravenously administered drug, as Applicants have demonstrated in the above-referenced application. This was even more surprising given the very low solubility and pH dependence of arsenic trioxide in water.

20. Moreover, one of ordinary skill in the art would not have been able to predict the greatly improved cardiac safety profile of orally administered arsenic trioxide compared to intravenously administered arsenic trioxide in view of the references cited by the Examiner, as demonstrated by the data herein.

21. We declare that all statements made herein of our own knowledge and belief are true and that all statements made on information and belief are believed to be true, and further, that the statements are made with the knowledge that willful false statements are

punishable by fine or imprisonment, or both, under section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

Date: 19-7-2007

Cyrus Rustam Kumana

Date: 25/7/2007

Yok-Lam Kwong